SALISBURY (J. H.)

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Hon Stephen Salisbury
with the kind regards & thanks
of the author –

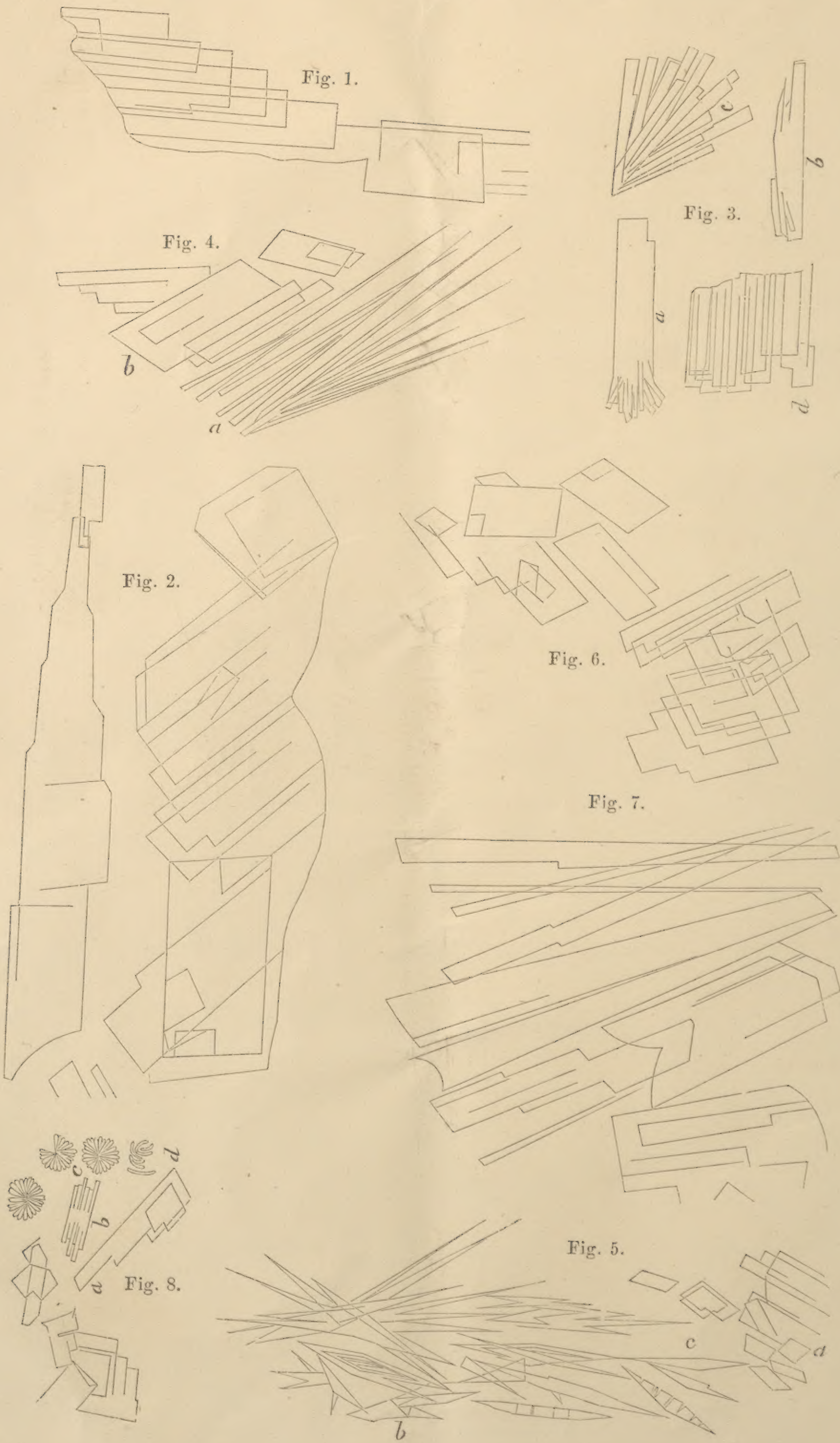
Fig. 1.
Fig. 3.
c
b
a
d
Fig. 4.
b
a
Fig. 2.
Fig. 6.
Fig. 7.
Fig. 8.
d
c
b
a
Fig. 5.
c
d
b

# PLATE II.

Fig. 10.

Fig. 9.

Fig. 11.

Fig. 13.

Fig. 14.

Fig. 17.

Fig. 12.

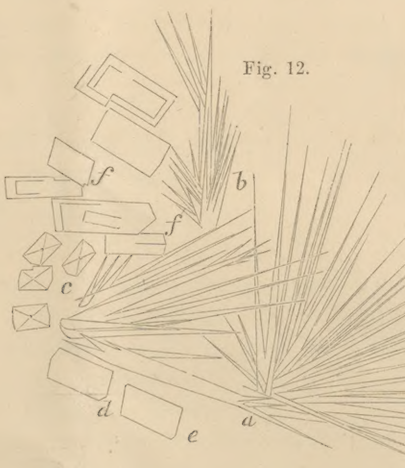


Fig. 15.

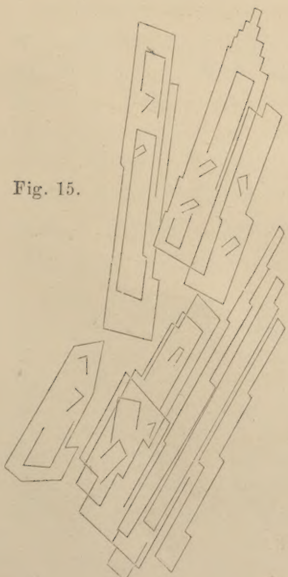

Fig. 16.

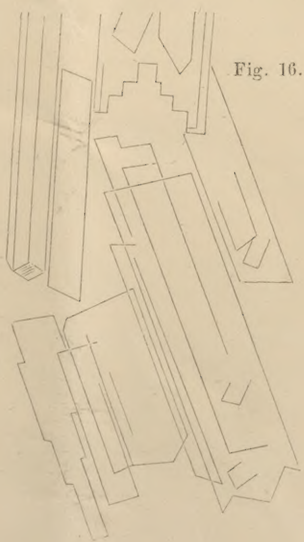

Fig. 18.

[Extracted from The American Journal of the Medical Sciences for April, 1863.]



# EXPERIMENTS

CONNECTED WITH THE

*Discovery of Cholesterine and Seroline, as Secretions, in Health, of the Salivary, Tear, Mammary, and Sudorific Glands; of the Testis and Ovary; of the Kidneys in Hepatic Derangements; of Mucous Membranes when congested and inflamed; and in the Fluid of Ascites and that of Spina Bifida.*

By J. H. SALISBURY, M. D.

(With two plates containing eighteen figures.)

I HAVE read, with much interest, the able paper of Austin Flint, Jr., M. D., of New York, entitled "Experimental Researches into a New Excretory Function of the Liver; consisting in the Removal of Cholesterine from the Blood, and its discharge from the body in the form of Stercorine;" published in the number of this Journal for October last.

Having in similar experiments, in which I have been for some time engaged, determined the presence of cholesterine and seroline (stercorine of Dr. Flint) in other healthy secretions of the human body—in the testis and ova of the human subject—in the ova of some animals—and in morbid collections of fluid in the human body, I here present briefly results which, so far as I know, are new.

The process followed for separating these bodies was, in all essential particulars, similar to that pursued by Dr. Flint, and for which I refer to his able paper.

*Exp.* 1. *Cholesterine in human ova.*—An unmarried woman, aged about 26, of strong constitution, was found dead in bed. On making a post-mortem examination, the stomach was found highly inflamed and ruptured. A gravid uterus was found, from which a fœtus, about three months gone, had recently been expelled. Near the body were found two bottles, one containing chloroform and the other oil of savin. The genital organs, stomach, and intestines, were taken for examination. Oil of savin was found in the fecal matter of intestines and in the contents of the stomach, which had been emptied into the peritoneal cavity. From all the

facts that could be gathered, from the post-mortem and chemical examinations, it appeared that the woman came to her death from taking oil of savin to produce abortion. The abortion was effected; but so much inflammation and softening of the stomach had been produced by the oil of savin that, during the severe vomiting the stomach was ruptured and death ensued. The ovaries were carefully washed and macerated in several waters, to free them from all adhering blood and serous matter; the ovisacs were then one by one punctured and their contents received into a porcelain capsule; this matter was evaporated to dryness over a water-bath, and the residuum subjected to the process for obtaining cholesterine and seroline. A concentrated hot alcoholic solution of the matter obtained was placed between glass slides to gradually crystallize. In a short time there appeared a beautiful mass of crystalline plates, which proved to be cholesterine, a portion of which is seen at Fig. 9. A quantitative examination was not made, but the qualitative revealed cholesterine largely present. No crystals of seroline appeared.

*Exp.* 2. *Cholesterine in ova of swine.*—The ovaries of pigs about six months old were taken and treated to separate adhering blood and outside serous matter; the ovisacs punctured, and their contents received into a large watch-glass capsule; evaporated to dryness over a water-bath, and residuum subjected to the process for obtaining cholesterine. The result was an abundant crop of crystals, having very much the appearance of those seen at Fig. 9, before referred to.

*Exps.* 3, 4, *and* 5. *Cholesterine in Graafian vesicles, liquor amnii, and brain of sow.*—A sow, weighing 525 lbs., was killed Dec. 1. She was about two years old, and very fat. The uterus contained twelve pigs that had attained about one-fourth their fœtal growth. Removed for examination, while the hog was still warm, the following parts: brain, contents of gall-bladder, liquor amnii, and ovaries. The brain (cerebrum and cerebellum) in its warm, fresh state, weighed 3147.59 grains. The gall-bladder contained of dark amber-colored bile 2160 grains. Bile alkaline, slightly ropy, and about the consistence of sweet oil at 90° F. Obtained from the ovisacs or Graafian vesicles between forty and fifty drops of a thin albuminous fluid, resembling the white of a hen's egg in feel and in coagulating properties. Of liquor amnii there was about four ounces to each pig. The amniotic fluid had a slight straw-yellow colour, was clear, and almost as limpid as water. It had an alkaline reaction, and the odor of fresh serum.

Subjected forty drops of the fluid from the Graafian vesicles to the process for determining the presence of cholesterine. Obtained a small crop of well-defined crystals resembling those seen at Fig. 9.

The liquor amnii, on cooling and standing in a porcelain evaporating dish for about six hours, contained on its surface a thin iridescent pellicle fissured by several sets of cleavage lines, which were often parallel to each other, and the different sets crossing either at acute or right angles. This pellicle proved to be cholesterine.

300 grains of the fresh brain (about equal parts of cerebrum and cerebellum, gray and white matter) were taken and subjected to the process for obtaining the cholesterine. The result weighed 3.897 grains. Percentage of cholesterine in fresh brain 1.299. The cholesterine as obtained had a beautiful white pearly lustre and crystalline appearance to the unaided eye. Redissolved in ether and allowed to crystallize slowly between glass slides, the crystals under the microscope had the appearance seen at Fig. 10.

*Exp.* 6. *Cholesterine in bile of sow.*—Subjected 800 grains of the fresh bile from the sow to the process for obtaining the cholesterine. The result weighed 5.614 grains. Percentage of cholesterine .702. The cholesterine had a white pearly lustre and crystalline appearance. Redissolved in ether and allowed to crystallize slowly between glass slides. The crystals had the appearance seen at Fig. 7. Many of the plates were very long and narrow.

*Exp.* 7. *Ova of hen.*—About half of the yolk of a fresh, newly-laid hen's egg was evaporated to dryness carefully over a water-bath, and the residuum proceeded with for obtaining cholesterine and seroline. The result was an abundant crop of crystals between the glass slides, a portion of which is seen at Fig. 1. The crystals were large and less rhomboidal than in the previous experiments. No seroline was found.

*Exp.* 8. *Ova of black bass.*—About one ounce of the ova of a large black bass (weight about eight lbs.) was examined in the usual way for cholesterine and seroline. After the final concentrated hot alcoholic solution had evaporated between glass slides, it was examined microscopically and found to contain a beautiful and abundant crop of cholesterine, a portion of which is seen at Fig. 2. The crystals were very large, and more rectangular than those from the human ova. No seroline was found.

*Exp.* 9. In the ovisac of the black bass described in the previous experiment were two large ragged calculi, one $3\frac{1}{2}$ inches long and $1\frac{1}{3}$ inches in diameter. They were covered with an iridescent membrane, resembling in appearance the metallic iridescence on the surface of the fish. These calculi were examined for cholesterine and seroline, and were found to contain the former in much larger quantity than the ova. The crystals had the same appearance as those seen at Fig. 2. No seroline was found.

*Exp.* 10. *Seminal fluid.*—Procured from a strong healthy man, aged about 35, two drachms of seminal fluid. Evaporated to dryness over a water-bath and proceeded with the residuum in the usual way for obtaining cholesterine and seroline. The final alcoholic solution, on evaporating between glass slides, was found to be composed of an abundant crop of crystals, a sample of which is seen at fig. 5. The amount of cholesterine was large, considering the quantity of material operated upon, but the crystals were small, as seen at *a;* a large proportion of the crystalline matter, however, was seroline, having the appearance of the crystals seen at *b* and *c*. Some of these crystals are seen to be rhomboidal, very acutely so, as at *c*.

*Exp.* 11. *Saliva.*—Evaporated to dryness and examined for cholesterine and seroline, two ounces of healthy saliva. Evaporated it over a water-bath in a broad, flat-bottomed porcelain dish; so that in thirty minutes after it was secreted the evaporation was complete. The saliva was from a strong healthy man, in the prime of life, who neither indulged in chewing or smoking tobacco, or in the drinking of intoxicating liquors. On placing the final alcoholic solution between glass slides, there was soon produced a large and beautiful crop of cholesterine crystals, the appearance of a portion of which is seen at Fig. 6. A quantitative examination was not made, but the qualitative analysis demonstrated it largely present in the saliva; apparently almost as abundant in this secretion as in the bile. No crystals of seroline were found.

*Exp.* 12. *Secretion from congested and inflamed mucous membrane.*—A strong healthy man contracted a severe cold from sleeping in a damp room. About forty-eight hours after, the mucous membrane of the fauces and nose became so congested and inflamed that a thin, watery, transparent fluid flowed from the nostrils at the rate of about two drops per minute. About one ounce of this fluid was collected. Under the microscope each drop was found to contain about 200 mucus cells and 100 colourless blood-disks. The one ounce was carefully evaporated to dryness over a water-bath and the residuum treated in the usual way for obtaining cholesterine and seroline. The presence of cholesterine was demonstrated in small quantity, much less than in the serum of the blood.

*Exp.* 13. *Bronchial mucus.*—Took two ounces of bronchial mucus expectorated by a patient labouring under severe chronic bronchial disease,[1] and subjected it to the process for determining the presence of cholesterine and seroline. A very small quantity of cholesterine was obtained, the appearance of which, as crystallized between the glass slides, is seen at Fig. 8, *a* and *b*. The crystal at *b* is split at the ends, like the crystals of cholesterine found in the fluid of ascites, seen at Fig. 3, *a* and *b*. The same form of crystal also occurs in jaundice urine. There was also found a small quantity of the peculiar crystalline matter seen at *c*, Fig. 8, and the bent prisms seen at *d*. From the small quantity of cholesterine found, it is probable that what there was came mostly from the small amount of saliva which necessarily would become mixed with the bronchial mucus in its passage through the mouth. The crystalline matter seen at *c* and *d* did not occur in the saliva examined. It is fair, therefore, to infer that it belongs to the bronchial mucus. The bronchial mucus was very thick, ropy, and white, having the appearance of coagulated albumen. It was quite acid. No pus present.

*Exp.* 14. *Serous fluid.*—Examined for cholesterine and seroline the serous fluid from the peritoneal cavity in a case of ascites in a female aged 73. "Liver extensively diseased. Had abscess a year ago, which pointed and discharged copiously at umbilicus. The dropsical effusion rapidly accumulated up to Nov. 12th, when she was tapped."[2]

The fluid had a milky appearance, and was thin and watery; slightly acid. Evaporated five ounces of the fluid to dryness over a water-bath, and subjected the residuum to the process for determining the presence of cholesterine and seroline. The final alcoholic solution gave, on being gradually evaporated between glass slides, some fine groups of crystals, samples of which are seen at Fig. 3, *a*, *b*, *c*, *d*. Those at *d* represent the usual form of cholesterine crystals; several groups presented the narrow radiating tablets seen at *c*, and still others had the appearance of those at *a* and *b*, the extremities of the plates being divided and bent.

*Exp.* 15. *Fluid of spina bifida tumour.*—Three ounces of fluid[3] from a spina bifida tumour in a child, was evaporated to dryness carefully over

[1] This patient was attended by Dr. Boerstler, to whose kindness I am indebted for furnishing the expectorated mucus for examination.

[2] I am indebted to the kindness of Dr. Effinger, of Lancaster, O., for the report of this case, and the fluid furnished for examination.

[3] Kindly furnished by Dr. Boerstler, of Lancaster, O., to whom I am indebted for many similar favours.

a water-bath, and treated further in the usual way for discovering cholesterine and seroline. The fluid was slightly alkaline. Cholesterine was demonstrated to be present in considerable quantity.

A portion of the fluid was examined carefully under the microscope. It was thin, watery, and milky, with numerous white flocks floating though it. These flocks under a low power (200 diameters) presented the appearance of fragments of milk curd. Under a higher power (600 diameters) they were resolved into the bodies mentioned below. Many of the "milk curd masses" were composed of nerve tubuli. Occasionally was found a tuberculated cell filled with fluid, and met with in most animal and vegetable tissues, and resembling somewhat some varieties of pollen grains. There were also found numerous spherical fungoid spores, aggregated in flocks, and among them was noticed a single elongated vegetating spore. Scattered through the fluid, quite abundantly, were flattish oblate spheroidal, highly refractive cells of various sizes, many of the larger of which had the appearance of being ruptured on the side. Occasionally one was met with containing small reddish-brown spherical cells.

In the flocks mentioned above were many remains of disintegrating cells. There were also numerous minute linear bodies, moniliform in structure, all through the liquid, and which in many instances were in active motion, making the whole mass of the liquid alive with them, the fluid being still fresh and sweet. These are the *so-called* "*vibriones;*" they have, however, no connection whatever with the *vibrii*, either in structure or mode of development. From the numerous examinations made, connected with diseased tissues, and fermenting and decaying bodies, I am strongly impressed with the belief that many of them are organisms, allied to sperm cells, being spermatozoid or antherozoid in character. There is evidence for believing some of them to be embryonic states of filamentous organisms belonging, or closely allied to, the confervaceæ. These latter are found in their mature state abundantly in the spleen, kidneys, and liver of animals and of the human subject, wound together in various ways and firmly attached to the glandular vessels. Analogous bodies are found in all stagnant waters, in the asci of Cryptogams, in the pollen grains of Phænogams, in the secretions and blood of the human body in all low typhoid types of disease, in all fermenting bodies, and especially during the incipient stages of active putrefactive fermentation (decay) in animal tissues. By inoculating fresh meat with them its decay is accelerated. During that stage of decay in the dead human body when dissecting wounds are so dangerous, they are very abundant and active; at later stages of the decay, when wounds are less dangerous, they are less numerous, and either motionless or sluggish, or are developed into inactive filaments. May they not be the source of the dangerous typhoid conditions produced by dissecting wounds? Occasionally was met a cell containing the sperm cells (so called vibriones) just described. Torula cells were frequently met with, either single or aggregated in masses. There were also several asci and numerous highly refractive sporoid bodies noticed, besides a peculiar barbed filament that I have frequently met with, connected with the glands and circulating fluids of animals.

*Exp.* 16. *Tears.*—Evaporated to dryness forty drops of freshly secreted tears, and treated the residuum for determining the presence of cholesterine and seroline. The tears were secreted by a lady of middle age, of remarkable health and vigour of constitution.

The residuum in the forty drops was large. On allowing the final alcoholic solution to evaporate slowly between glass slides, a fine crop of cholesterine crystals was the result. They resembled much the crystals in Fig. 16.

*Exp.* 17. *Milk.*—Milk of a young married lady, of vigorous constitution, eight months gone with her first child. Evaporated one drachm to dryness carefully over a water bath—within a few minutes after it was drawn—and subjected the residuum (which was large) to the process for determining cholesterine and seroline. The final hot alcoholic solution, on evaporating between glass slides, yielded a large crop of rhomboidal and rectangular plates of cholesterine—a sample of which is seen at Fig. 11. No crystals of seroline were discovered. A quantitative analysis was not made; but the qualitative demonstrated cholesterine to be largely present.

*Exp.* 18. Milk from a lady of fine constitution, nursing her second child, which is nine months old. Milk abundant, and child healthy and robust. Took 500 grains of the newly secreted milk and evaporated it to dryness over a water bath, and proceeded with the residuum (which was large) to determine the presence and amount of cholesterine and seroline. On evaporating the final alcoholic extract to dryness carefully over a water bath in a tarred porcelain capsule, I found .64 of a grain of unsaponifiable matter, which proved to be seroline and cholesterine. From the apparent relative proportion of the crystals exhibited between the glass slides, I should estimate over one-half to be cholesterine.

The result may be stated as follows:—

| | | |
|---|---|---|
| Freshly secreted milk . . . . . . | 500 | grains. |
| Stercorine and cholesterine . . . . . . | .64 | " |
| Percentage of stercorine and cholesterine . . | .128 | " |

Fearing there might be some error in this result I repeated the process—carrying on the saponification with a strong solution of pure caustic potassa—at a temperature of 212° F. over a water bath, for one and a half hours, repeatedly stirring; then largely diluting with water and filtering, &c. The result was the same as in the preceding experiment. A concentrated hot alcoholic solution, placed between glass slides, crystallized in long slender, often radiating needles, as seen in Fig. 12. Mixed with these crystals of seroline, appeared plates of cholesterine, *c*, *d*, *w*, *f*. These crystals first appeared as seen at *c*, then passed into the forms *d*, *d*, and these into the forms *w*; and *w* into the forms seen at *f*, *f*.

*Exp.* 19. Took 700 grains of healthy, freshly secreted cow's milk (about five months after coming in), evaporated to dryness over a water bath, and treated the residuum as hereafter described for obtaining seroline and cholesterine. The final alcoholic solution, being carefully evaporated and thoroughly dried over a water bath, in a tarred porcelain capsule—weighed .759 of a grain, which proved to be seroline and cholesterine. On redissolving in hot alcohol and allowing a concentrated solution to crystallize between glass slides, the seroline crystals were found to resemble those from the milk of the human subject (Fig. 12). Those of cholesterine were rather peculiar.

The result of the analysis may be stated as follows:—

| | | |
|---|---|---|
| Fresh cow's milk . . . . . . . | 700 | grains. |
| Seroline and cholesterine . . . . . . | .759 | " |
| Percentage of seroline and cholesterine . . | .1084 | " |

The last portions dissolved from the dry matter of the milk, by ether, contained the majority of the cholesterine. The seroline and the saponifiable fats were the first to be dissolved. For this reason, two ethereal extracts were made from the dried milk. In the first, the dried pulverized matter was digested at a temperature of 75° to 80° F. for twelve hours, in pure ether, frequently stirring, the vessel being so covered as to prevent as much as possible the evaporation of the ether. It was then filtered, and this first extract proceeded with in the usual way. No cholesterine was detected in it; but quite a large percentage of seroline, which in its crystals resembled that of the human milk (Fig. 12). The dry matter was a second time digested in ether, frequently stirring, for twenty-four hours; then filtered, thoroughly washing with warm ether, and this second extract proceeded with as with the first. The final alcoholic extract, on being evaporated to dryness over a water bath, left a yellowish matter having something the appearance and consistence of unbleached wax. Its lustre, however, was more pearly. It was but slightly soluble in cold alcohol; slowly soluble in hot alcohol, and readily soluble in ether both cold and warm. The concentrated ethereal solution, on being placed between glass slides, so as to prevent its too rapid evaporation, soon crystallized into beautiful cholesterine plates.

There were a few acicular prisms or plates, bent in the form of a crescent. They resembled those seen at *d*, Fig. 8.

Milk holds cholesterine with great tenacity, and requires long digestion to completely separate it. The early secreted milk in the human subject, previous to birth, is quite free from seroline; but after birth, and during nursing, the seroline is secreted largely with the cholesterine.

*Exp.* 20. *Intermittent fever. Perspiration.*—Sept. 10. John Laudabagh[1] was attacked with intermittent fever. Type quotidian. Had been free from the disease for eighteen years previous. On the afternoon of the 14th (fifth day of disease) procured about one-half ounce of the perspiration, during the sweating stage of the paroxysm. Perspiration alkaline, limpid like water, and perfectly clear and transparent. Subjected two drachms to the process for obtaining cholesterine and seroline. Obtained a beautiful crop of crystals, a sample of which is seen at Fig. 4, *a* and *b*. The seroline and cholesterine appeared to be about equally divided. The seroline crystals were very beautiful, long, slender needles. When a crystal was alone, so that the whole of it could be seen, one end presented the appearance of a narrow rectangular plate, while the other tapered off into a sharp acicular point. Sometimes the bases (large ends) of the crystals were bifurcated, and at others the apices were more or less divided. The cholesterine crystallized in large rhomboidal and rectangular plates. A quantitative examination was not made, but the qualitative determined these bodies to be largely present in the perspiration of intermittent fever.

*Exp.* 21.[2] *Urine.*—Charles Whitney, aged five years, was attacked Sept. 13, with intermittent fever—quotidian type. Never had chills and fever before. He resides in the 3d ward near and but a few feet above the ague bogs. At 11 A. M., Sept. 13, was taken with the first well-marked paroxysm. The sweating stage terminated at 4 P. M. On the following

[1] This case was reported and perspiration furnished by Dr. Effinger.

[2] I am indebted to the kindness of Dr. Effinger for the reports and the materials furnished for examination of the cases in Experiments 21 to 33, and 35 and 37.

day (Sept. 14) the paroxysm commenced at 9 A. M. The algid, pyrexial and sweating stages were all well-marked. Voided the sample of urine—of this experiment—at 3 P. M., as the febrile stage was passing off and the sweating coming on.

Subjected two ounces of the urine to the process for obtaining cholesterine and seroline. Obtained from the final alcoholic solution a large crop of beautiful rhomboidal and rectangular plates, a sample of which is seen at Fig. 18. Some of the crystals were divided at the extremities, as is seen at *a*.

*Exp.* 22. Mr. C——, aged 48, was attacked with intermittent fever, tertian type, Sept. 1. Arrested after the second paroxysm. Relapse Sept. 15. Arrested the second time after the second paroxysm. Urine obtained Sept. 21, four days after the arrest of the paroxysm the second time. His complexion was sallow, appetite poor, with considerable physical prostration, and mental lethargy.

His residence was near a large excavation which was being made in the low ground adjoining the canal, and from which he was constantly exposed to the seeds of the disease. Never had the disease before.

One ounce of the urine was subjected to the process for determining the presence of cholesterine and seroline. Well-marked rhomboidal and rectangular plates of cholesterine were obtained, resembling those at Fig. 18. No seroline found.

*Exp.* 23. James Scott, aged 13 years, living immediately on the border of the ague bogs, at Lancaster, O., and but about five feet above them, had been labouring under intermittent fever most of the time since the 1st of August. Type quotidian. Obtained his urine Sept. 26, during the interval between the paroxysms, and two days after he had commenced taking quinia (previously he had been dosed with herb teas). Subjected one ounce of the urine to the process for determining the presence of cholesterine and seroline. Obtained a few small well-marked crystals of the former body, but none of the latter.

*Exp.* 24. Ellisworth McLean, aged 17 months, was attacked with intermittent fever — quotidian type — during the forepart of August. Dr. Effinger saw the child for the first time on the 7th of October. He reports him as much reduced, very pale, flesh doughy; face bloated, and feet and legs œdematous. Spleen enlarged, forming a well-marked "ague cake." He commenced his treatment with acetate of potash. He obtained the urine for me on the 9th of October, during the apyrexial stage. Urine pale, clear, and slightly alkaline. Subjected two ounces to the process for determining the presence of cholesterine. Obtained a fine crop of well-marked crystals, resembling those at Fig. 18.

*Exp.* 25. *Urine of remittent fever.*—Mr. C——, aged about 30, had been labouring for some days under an attack of remittent fever. Obtained his urine on the ninth day of the disease, when convalescing. Treated two ounces of the urine for cholesterine and seroline. Obtained a fine crop of cholesterine crystals, with which were mixed a few crystals of seroline.

*Exp.* 26. *Urine of typhoid fever.* — Mrs. E——, a married lady of about 30, of good constitution, was attacked with typhoid fever, August 26. On Sept. 9, the fourteenth day of disease, obtained his urine. Urine acid, high-coloured; full of active "vibriones," and had a large deposit of

rhomboidal and lozenge-shaped prisms and plates of lithic acid. Pulse from 100 to 120. Considerable enteric tenderness, with slight diarrhœa. Subjected two ounces of the urine to the process for determining the presence of cholesterine and seroline. Obtained a large crop of well-defined rhomboidal and rectangular plates, a sample of which is seen at Fig. 16. The crystals were formed between glass slides, and were very large and beautiful. At *a* is a long 6-sided prism, made up of distinct laminæ.

*Exp.* 27. Mrs. E—— (the same case as 26), on the 18th of September and the twenty-second day of the disease, appeared to be improving. The mind was clearer, and appetite better; but the pulse still remained up to 110 and 120, and was small. Skin dry. Strength of body and mind improved since Sept. 9th. Obtained her urine. It was lighter in colour and contained less sediment than the previous one. The character of the sediment was, however, the same. Subjected two ounces to the usual process for obtaining cholesterine. Obtained a fine crop of crystals like those at Fig. 16.

*Exp.* 28. Mrs. E—— (same case as 26 and 27)—typhoid fever—Oct. 20, fifty-fourth day of disease. Patient still in a very low, depressed state. Pulse 120, but no fever; is very pale, body feeble, mind weak, nervous, and fretted by the least noise. Hearing completely restored and very sensitive. Has scarcely any appetite, sleep nervous and disturbed. Urinates frequently and quite copiously. Obtained the urine Oct. 20, through the kindness of her attending physician, Dr. Effinger. Urine pale (still containing many vibriones), strongly acid, and contains much less sediment than the previous samples.

Subjected two ounces to the process for determining the presence of cholesterine and seroline. Found cholesterine still present, in considerable quantity; quite as large as in the previous examinations. The plates had the appearance of those at Fig. 16. No seroline was found.

*Exp.* 29. Libby ——, domestic at Mrs. E.'s, aged 25, down with typhoid fever. Attended Mrs. E. in her attack of typhoid fever. Libby was attacked on the 4th of October, thirty-eight days after Mrs. E. was taken down. Oct. 20, the seventeenth day of the disease, obtained her urine through Dr. Effiinger, her attending physician. He reports the diarrhœa constant, and the most annoying symptom, indicating enteric glandular depositions and inflammation. For the last week the passages were mixed with blood. Mind yet clear, but very deaf; has been so for over a week. Prostration not very great, less than ordinary. Urine slightly acid, high-coloured, with considerable lithic sediment.

Subjected two ounces of the urine to the process for obtaining cholesterine and seroline. Obtained a fine crop of crystals of cholesterine, which resembled those seen at Fig. 16. No seroline found.

*Exp.* 30. Libby (same case as the above, at an earlier date), Oct. 8, fourth day of the disease. Obtained urine, and subjected it to the process for obtaining cholesterine and seroline. Obtained well-defined rhomboidal and rectangular plates of cholesterine. The amount present was less than in the urine of the preceding experiment. No seroline found.

*Exp.* 31. Samuel E——, aged 11 years, son of Mrs. E——, was taken down with typhoid fever on the 8th of October. On October 20th—the twelfth day of the disease—obtained his urine, through the attending phy-

sician, Dr. Effinger. He reports the case a mild type of the disease. He says, "There are no marked symptoms of disturbance of the system, either functional or organic, except at night, when he is restless and feverish. Though an active and sprightly boy, he has no disposition to set up or leave his bed. While there recumbent he is cheerful and pleasant, amusing himself with his slate and books. As soon as he gets up and moves about he becomes pale and sick, and goes back to bed."

Urine strongly acid, of a yellowish colour, with considerable flocculent sediment. The upper portion of the precipitate is light coloured, and the lower of the colour and appearance of brick dust. This lower portion is made up of lithates. Two ounces of the urine were subjected to the process for obtaining cholesterine and seroline. A few well-defined rhomboidal and lozenge-shaped tablets of cholesterine were obtained. The cholesterine was small in this urine. No seroline.

*Exp.* 32. Lucy ——, the sister and nurse of Libby (domestic to Mrs. E——), was attacked with typhoid fever on the 8th of November, thirty-six days after her sister Libby was attacked, and whom she attended as nurse. Obtained her urine, through the attending physician, Dr. Effinger, on the 22d day of November, the fourteenth day of the disease. "Fever constant, but not high. Diarrhœa set in on the 20th November (day before yesterday). Discharges frequent and bloody. Urine acid, high coloured, and contains a large precipitate of lithates.

Two ounces of the urine were subjected to the process for obtaining cholesterine and seroline. A fine crop of cholesterine crystals was obtained, the appearance of which were like those in Fig. 16. Crystallized on the glass slides it had a beautiful pearly lustre to the naked eye. Cholesterine occurred largely in this sample of urine. No seroline was found.

*Exp.* 33. *Urine of diphtheria.*—Lizzie Prentice, aged 5 years, labouring under well-marked diphtheria. Morning urine obtained, and two ounces subjected to the process for obtaining cholesterine and seroline. Obtained a few well-marked crystals of cholesterine. No seroline.

*Exp.* 34. *Urine of varicella.*—Sallie Mattock, aged 8 years, labouring under well-marked varicella. Her morning urine was obtained when the eruption was at its height. Treated two ounces of it for cholesterine and seroline. Obtained well-marked crystals of cholesterine, a sample of which is seen at Fig. 17. No seroline found.

*Exp.* 35. *Urine of jaundice.*—Urine of a patient labouring under well-marked jaundice Urine voided on the fifteenth day of the disease. Urine slightly acid, high coloured, with quite a large flocculent yellow precipitate. Two ounces of the urine were evaporated over a water-bath to dryness, and the residuum treated in the usual way for cholesterine and seroline. The final alcoholic extract, on being placed between glass slides and allowed to stand for six hours, gave a fine crop of crystals; some of which represent the usual crystalline form of cholesterine. The crystals were small, but quite numerous. Some of the crystals were split at the ends, like those at *a* and *b*, Fig. 3. Others were very acutely rhomboidal; while others were acicular, yet retained somewhat the very acutely rhomboidal outline, where they were perfect and alone. These last were undoubtedly crystals of seroline, and were quite abundant.

*Exp.* 36. Urine from a lady affected with jaundice. Urine voided during

the third week of the disease. Had a high colour, strong odour, and a small, yellowish sediment, resembling oil globules. Treated in the usual way—one ounce for cholesterine and seroline. Obtained a very fine crop of crystals, consisting of long rhomboidal and rectangular plates of cholesterine, with which were mixed a few acicular crystals of seroline.

*Exp.* 37. Mrs. M——, cook on board a canal boat, aged 53, was attacked with jaundice about the first of September, and is still labouring under the disease (October 26th). Obtained urine, through her attending physician, Dr. Effinger, and subjected two ounces to the process for determining the presence of cholesterine and seroline. The final alcoholic solution, evaporated between glass slides, afforded a fine crop of cholesterine crystals, showing this body quite largely present.

*Exps.* 38, 39, and 40—*Butter, and beef and hog suet*—were the examinations of butter, and beef and hog suet. Cholesterine and seroline were demonstrated in all of them. In beef suet, cholesterine occurs quite largely.

*Exp.* 41. *Urine of diabetes mellitus.*—Mr. ——[1], labouring under a severe attack of diabetes. The patient is a robust, middle-aged man, who has, previous to this attack, enjoyed good health. He passed about 192 ounces of urine daily. It was rich in sugar, and underwent active fermentation ; during which the torula (yeast) cells and filaments were greatly multiplied. A sample of the urine ($\frac{1}{2}$ pint), the next day after it was voided, was subjected to the process for determining the presence of cholesterine and seroline. From the $\frac{1}{2}$ pint, 4.32 grains of cholesterine were obtained. The crystals were very large and beautiful, a sample of which is seen at Fig. 15. No seroline was obtained. From this experiment it is seen that cholesterine occurs largely in diabetic urine. The discharge of a single day (192 ounces) would contain 103.68 grains of cholesterine. The discovery of cholesterine, as a secretion of the kidneys in *diabetes mellitus*, may throw some additional light upon the nature of the disease, and perhaps suggest some modifications in its treatment.

*Exp.* 42. *Healthy urine.*—Examined 8 ounces of healthy urine for cholesterine and seroline. The urine was voided by a pregnant lady, of fine constitution, eight months gone with her first child. Urine slightly acid. Was unable to detect the least trace of either of the bodies sought for.

*Exp.* 43.—Examined the urine of a strong, healthy man, aged about 30, for cholesterine and seroline. Subjected 4 ounces to the usual process (the one used in the preceding experiments), but was not able to detect a trace of either of the bodies.

*Exp.* 44. *Perspiration in health.*—Examined 1 drachm of healthy perspiration (secretion excited by vigorous exercise) for cholesterine and seroline. Well-marked crystals of cholesterine were found. No seroline.

*Exp.* 45. *Ovarian tumour.*—The following note from Dr. Effinger, of Lancaster, Ohio, explains the subject of this experiment :—

"Dear Doctor: Yesterday morning I made a *post-mortem* of Mrs. Calvin Tripp, who died of ovarian tumour. The tumour weighed 62 pounds, water 15 lbs., and hard part of tumour 47 lbs. I made a few slices from different parts

[1] I am indebted to Dr. Boerstler for reporting this case and furnishing the urine for examination.

of the tumour, which I send you. I found imbedded in the substance a small, dead worm that may possibly be a novelty. You will find it in the cut I made near its burrow cell.

"Respectfully yours,
"M. EFFINGER.

"LANCASTER, Nov. 29th, 1862."

The tumour had a white, fatty appearance, to the unaided eye. It was, however, very firm and tenacious. The microscope demonstrated it to be strictly a fibrous tumour. A portion of it was examined for cholesterine and seroline, but neither of them were detected.

Fig. 13 represents the worm referred to in the above note of Dr. Effinger. Length two lines; diameter $\frac{2}{5}$ of a line. To the naked eye it had the appearance of a small fragment of clotted blood. Viewed with a $\frac{1}{4}$ inch objective, it appeared the size seen at Fig. 13. Colour blood-red, except the sucker and around the margins of the body. When emptied of its food it had a transparent white colour. It had one sucker, and that on the anterior part of the body. It appears to be allied to the genus Festucaria.

While examining a thin slice of the tumour under the microscope, I discovered among the fibres, the mite represented at Fig. 14. Length $\frac{1}{300}$ of an inch. A short time since, while examining the perspiration from a patient labouring under intermittent fever, I found a different species of the same genus.

*Chemical Properties of Cholesterine.*—The sample of cholesterine, obtained from the second ethereal extract from milk, remained unfused at a heat considerably above that of boiling water. Its precise freezing point was not determined. The freezing point of cholesterine is stated by different authorities at 279° and 293° F.

Concentrated $SO_3$ strikes a beautiful purple red colour, with the cholesterine obtained in the foregoing experiments. In order to see this test to advantage, a little cholesterine should be spread thinly on a glass slide or plate, and a drop of concentrated $SO_3$ placed on and spread over it by means of a glass rod. The beautiful purple red tint will begin to show itself—first, around the edges of the acid, and, in a few minutes, the whole surface will assume this beautiful colour. Heat should not be applied, nor too much acid used.

Cholesterine is soluble in 9 parts of boiling alcohol of 0.84, and 5.55 parts of 0.816. It is soluble in 12 parts of ether at 32° F., 3.7 at 59° F., and 2.2 at boiling. It is also soluble in wood-spirit, slightly soluble in boiling oil of turpentine and in water containing 4 parts of dry soap. Heated with $SO_3$ it decomposes, and $NO_5$ changes it into cholesteric acid and artificial tannin. Form. $C_{38}H_{33}O$, or $C_{36}H_{32}O$.

*Chemical Properties of Seroline.*—Lehman gives the melting point of seroline at 98° 8′ F. Boudet at 97° F.

Boudet's process for obtaining seroline from the blood—was to evaporate the blood to dryness, treat the residuum thoroughly with water, then dry completely the residuum, and repeatedly treat it with boiling alcohol.

This alcoholic extract on cooling let fall the seroline in flocks. By this process Boudet must have obtained with the seroline some small portions of margarine and stearine, which would have a tendency to elevate the melting point.

For obtaining the seroline, which was used for determining the fusing point of this body, I evaporated to dryness over a water bath, pulverized in an agate mortar and digested with pure ether for twelve hours—repeatedly stirring and keeping the vessel so covered as to prevent the evaporation of the ether—filtered, evaporated filtrate to dryness over a water bath, and digested the residue at 212° with a strong solution of pure caustic potassa for one and a half hours to saponify the saponifiable fats; then largely diluted with water and filtered, washing the filter with water till the fluid came through neutral. The filter was then thoroughly, yet carefully dried over a water bath, placed in a warm covered funnel and kept filled with ether till that which passed through, on evaporation, left no residuum. The filtrate was then evaporated to dryness over a water bath and redissolved in hot absolute alcohol, and the seroline allowed to deposit as the alcohol cooled and evaporated spontaneously. Before the seroline was tested for the fusing point, it was exposed to the heat of 212° over a water bath in a watch glass, until completely deprived of all alcohol and moisture. It was then allowed to cool and crystallize at a temperature of 50° F. When perfectly crystallized, its fusing point was determined.

The seroline thus obtained began to lose its white colour and crystalline structure at 90° F. At 94° F. it had the consistence of Canada balsam and had lost entirely its crystalline structure, and at 96° F. it flowed readily, and was about the consistence of sweet oil. At this last temperature it had a light straw yellow tinge. At 32° F. it had a white pearly lustre, like spermaceti, but less consistent. At 50° F. it had the consistence of lard at 80°. When allowed to crystallize slowly, it crystallized in long slender needles, as seen at Figs. 12 and 4. Cold, it was odourless; at 212° it exhaled a sweet oily odour. Almost insoluble in cold alcohol, but very soluble in ether and hot alcohol. Strikes, like cholesterine, a beautiful violet red colour with strong $SO_3$.

*Primary Form of the Crystals of Cholesterine and Seroline.*—The primary form of the crystals of cholesterine appears to be the cube and rhombic prism. In a few instances, in the early stages of the crystallization, hexagonal prisms were noticed; but these were probably formed from the rhombic prisms by the truncation of the acute angles, as is the case in some crystals of mica. The octohedra, rectangular prisms and plates may be secondary forms of the cube and rhombic prism.

In these experiments there has appeared some evidence in favour of the very acute rhombic and rhomboidal prisms being the primary form of the acicular crystals of seroline.

*Resumé.*—The following is a brief summary of the facts indicated by the preceding experiments:—

1. Cholesterine occurs largely in the ova of the human subject and of animals.

2. In the seminal fluid of the human subject, seroline and cholesterine are largely present, the former more so than the latter.

3. Cholesterine occurs very largely as a secretion in the saliva. No seroline is found.

4. Neither seroline nor cholesterine occurs in healthy urine.

5. Cholesterine occurs quite largely, and seroline in small quantity in jaundice-urine. (These bodies are probably always secreted by the kidneys whenever the liver, through organic or functional derangements—is unable to secrete them from the blood.)

6. Cholesterine and colourless blood disks are secreted or effused from highly congested and inflamed mucous surfaces.

7. Cholesterine is secreted or effused from the peritoneal (serous) membrane in ascites.

8. Cholesterine occurs largely in the fluid of spina bifida tumours.

9. Cholesterine is secreted by the tear glands.

10. Human milk, previous to birth, is rich in cholesterine. No seroline detected in the experiment made.

11. After the birth of the child, and during nursing, the mammary glands secrete largely cholesterine and seroline.

12. The milk of the cow is rich in cholesterine and seroline.

13. Butter, beef, and hog suet contain cholesterine and seroline.

14. The primary forms of the crystals of cholesterine appear to be the cube and rhombic prism; and that of seroline, the very acute rhombic or rhomboidal prism; though usually appearing as simply acicular.

15. Cholesterine and seroline are largely secreted from the blood by the sudorific glands during the sweating stage of intermittent fever. These glands become important blood depurative organs in this disease.

16. The kidneys largely secrete cholesterine in intermittent fever.

17. The kidneys secrete cholesterine in varicella.

18. The kidneys secrete cholesterine in diphtheritic conditions.

19. The kidneys largely secrete cholesterine in the disease known as *diabetes mellitus*.

20. The kidneys secrete cholesterine and seroline in remittent fever.

21. The kidneys largely secrete cholesterine in typhoid fever.

22. Cholesterine is secreted by the sudorific glands in health.

*Concluding Remarks.*—Cholesterine appears to be essentially a body, secreted from the blood by the glands concerned in digestion; the sudorific glands; those secreting tears and milk; and by the testis of the male and ovary of the female, and by the kidneys in hepatic disease. In the secre-

tions of the testis (seminal fluid), seroline occurs more largely than cholesterine. In the female ova, cholesterine occurs largely, and no doubt has some office to perform in furnishing one important constituent of nourishment in the early fœtal development; before, in viviparous animals, there are any uterine attachments; and in oviparous, before they escape from the ovarian envelopes. Mucous and serous surfaces do not appear to have any power to separate cholesterine from the blood; unless perhaps when under the influence of congestion and inflammation.

As cholesterine occurs so largely in the bile and saliva, two secretions important in digestion, in the female ova, and in the mother's milk upon which the young feed, is it not highly probable that it has some important function to perform in digestion, at all ages; and as nourishment and a soporific in infancy, it only becoming excrementitious proper when this office is ended, and it is changed into seroline (stercorine of Dr. Flint)?

It is believed to be pretty well established, that the true source of cholesterine is the nervous system, of which it is an effete product. From the nervous system it passes to the blood, and is removed from the blood by the liver.

These experiments go to show that the liver is not the only organ which separates this body from the blood. The salivary, tear, mammary, and sudorific glands; and the testis and the ovary come in, each in its peculiar time and place, as important aids. They also show that a portion of the cholesterine of the human body may be taken into it through the food eaten, consisting of milk and butter, eggs, beef, and hog fat, and as there are more or less blood and serous matter in meat, be taken in in that substance also. Still, these facts do not argue against the nervous system being its true original source. They only show that it is formed in the nervous system of animals as well as in that of the human subject; and that in feeding upon animal food, the vascular system may gather this substance from two sources, the nervous system and the food eaten. The nervous system being the source of cholesterine, and the tear glands secreting this body, may explain why the profuse shedding of tears, in health, for any great length of time, so enervates both physically and mentally.

All functional and organic derangements of the liver produce despondency. The dark side of the picture is the one ever prominent. Actions and remarks are perverted, and everything goes wrong. There is a tendency for this condition to relieve itself, especially in the female sex, by a profuse flow of tears. May not this peculiar mental and moral condition, full of sad forebodings, be but a part of that beautiful sympathy of action between different organs of the body, wisely designed, in this instance, to stimulate the tear glands to excited action, in order that they may perform, to some extent, the depurative office of a liver, and thus relieve, partially, a vascular system surcharged with cholesterine?

That weeping relieves sad and despondent conditions is so true that you

everywhere find it proverbial; it is well known that sudden grief does much towards deranging the functions of the liver. The tear glands, through sympathy, appear to come in as little safety-valves to the vascular system on such occasions, as well as on others hereafter mentioned, where the liver is deranged in its functions.

In climates where there is a disposition to "biliousness" ("bilious climates"), there is a tendency to inaction of body and mind; a heavy lethargic feeling prevails; a greater tendency to lounge about lazily and to sleep than in less "bilious" localities; the intellect is inactive and heavy; there is also a tendency to the greater deposition of adeps—a tendency to obesity.

In all diseased conditions of the liver where its normal functions are impaired, there is great dulness and lethargy, with a feeling of melancholy sadness and a disposition often to doze and sleep.

Children while nursing sleep a great portion of the time; they fall asleep while feeding: there is also a remarkable tendency to take on fat. After being weaned they are much more wakeful, and the fatty deposits usually decrease.

The free use of cows' milk as food produces heaviness and a tendency to sleep. The use of eggs largely as food produces a similar lethargic condition.

May we not account for the lethargic influence and the tendency to sleep and obesity of "bilious climates," on the ground of the blood and nervous system becoming and remaining constantly surcharged with cholesterine? In diseased conditions of the liver, when its depurative functions are impaired, we know the blood and nervous system become surcharged with this body, and we know that this surcharged condition produces results similar to those of "bilious climates."

May not the cholesterine and seroline in the food of nursing infants be one cause of their disposition to sleep and to become fat?

May not these bodies also, in milk and eggs, be the cause of their producing heaviness and sleep? When using milk and eggs as food, the liver has to perform the double office of removing the cholesterine formed by the nervous system and that taken into the blood by the food eaten. The result is that the blood and nervous system become surcharged with this body, and we have temporarily the same condition of the system that occurs in "bilious climates." The liver being more or less deranged in its functions in intermittent fever, the sudorific glands come in as blood depurative aids in freeing the vascular system of cholesterine and seroline and other effete matters. This may explain, to some extent, the advantage derived from the free use of diaphoretics as aids in the successful treatment and eradication of this disease.

The secretion of cholesterine from the blood by the kidneys, in intermittent fever, may explain why it is that the free use of diuretics (acetate of potash, &c.) are so beneficial often in its treatment. Without the free use

of diaphoretics and diuretics in the treatment of intermittent fever the disease is seldom so perfectly eradicated from the system as to prevent its reappearance the following spring; while with their proper use, the disease seldom reappears unless the system is again exposed for some length of time to the exciting cause.

The discovery of the fact that the kidneys secrete cholesterine largely in diabetes mellitus may throw some light upon the pathology and therapeutics of this peculiar disease.

*Ready process for detecting the presence of cholesterine and seroline.*—As the ordinary process for determining the presence of cholesterine and seroline is too lengthy and often beyond the facilities of the practising physician, I here give a simple method, which may answer as a very good approximate means (till a better is suggested) for detecting these bodies in urine and other secretions, and which may often be used with advantage by physicians in their practice as a diagnostic aid. Care should be taken, in the microscopic examination of crystals obtained from the secretions by this process, to not confound those of cholesterine with those of lithic acid and chloride of sodium. As the secretion of these bodies by the kidneys is an abnormal function, one which they only perform when the liver, whose normal office is to secrete these bodies, is deranged, their presence in the urine will usually indicate hepatic derangement.

Place two to four ounces of urine in a six ounce bottle, and add one ounce of pure ether; tightly cork, and agitate by turning quickly the bottle on different ends, allowing the ether to pass backward and forward through the urine, so as to wash it completely—two to five minutes' stirring are usually sufficient;—then allow the ether to rise to the surface, and decant into a clean porcelain or glass dish and evaporate carefully to about ten drops, which place between glass slides and set aside for several hours to crystallize. When this is completed, a microscopic examination with a moderate power will detect the cholesterine plates and acicular crystals of seroline, if present.

In concluding, I take pleasure in expressing my obligations to Drs. Boerstler and Effinger, of Lancaster, Ohio, for valuable aid. They have felt a lively interest in these experiments, and have done me many favours in reporting well-marked cases of disease and in furnishing specimens for examination.